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<input type="checkbox"/>	L1	equine.clm. and neurona.clm.	16
<input type="checkbox"/>	L2	sarcocyst\$ near10 vaccin\$	18
<input type="checkbox"/>	L3	l2 not l1	13

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<input type="checkbox"/>	L3	l2 not l1	13
<input type="checkbox"/>	L4	(ng.in. or bigbie.in. or whalen.in.) and (sarococystis or neurona or merozoite)	8

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## Search Results - Record(s) 1 through 8 of 8 returned.

☐ 1. [20040110295](#). 28 May 03. 10 Jun 04. Nucleic acid vectors. Punnonen, Juha, et al. 435/455; 435/320.1 435/6 536/23.1 C12Q001/68 C07H021/04 C12N015/85.

☐ 2. [20040001849](#). 07 Mar 03. 01 Jan 04. Antigen library immunization. Punnonen, Juha, et al. 424/186.1; 424/188.1 424/189.1 424/190.1 530/350 A61K039/12 A61K039/21 A61K039/29 A61K039/02 C07K014/16 C07K014/02 C07K014/195.

☐ 3. [20020198162](#). 10 Feb 99. 26 Dec 02. ANTIGEN LIBRARY IMMUNIZATION. PUNNONEN, JUHA, et al. 514/44; A61K031/70 A01N043/04.

☐ 4. [20020041886](#). 23 Apr 01. 11 Apr 02. Equine protozoal myeloencephalitis vaccine. Bigbie, Rocky Barry, et al. 424/269.1; 435/258.1 514/44 A61K048/00 C12N001/10 A61K039/002.

☐ 5. [6576757](#). 28 Nov 00; 10 Jun 03. Polynucleotides encoding flavivirus and alphavirus multivalent antigenic polypeptides. Punnonen; Juha, et al. 536/23.72; 424/184.1 424/204.1 424/218.1 424/228.1 536/23.1. C07H021/04 A61K039/12 A01N043/04 .

☐ 6. [6569435](#). 28 Nov 00; 27 May 03. Flavivirus and alphavirus recombinant antigen libraries. Punnonen; Juha, et al. 424/202.1; 424/204.1 424/234.1 424/236.1 424/274.1 435/320.1 435/6 514/44. A61K039/12 A61K039/295 A01N043/04 .

☐ 7. [6541011](#). 10 Feb 99; 01 Apr 03. Antigen library immunization. Punnonen; Juha, et al. 424/204.1; 424/218.1 530/300 530/350. A61K039/12 C07K001/00 .

☐ 8. [US20020041886A](#). Vaccine useful for preventing or ameliorating equine protozoal myeloencephalitis disease, comprises inactivated *Sarcocystis neurona* cells and/or *Neospora hughesi* cells, antigens, DNA derived from the cells or their mixtures. BIGBIE, R B, et al. A61K039/00 A61K039/02 A61K039/395 A61K048/00 A61P033/00 C07K016/20 C12N001/10.

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Term	Documents
NG	1146264
NGS	71893
BIGBIE	90
BIGBIES	0
WHALEN	4081
WHALENS	0
SAROCOCYSTIS	0
SAROCOCYSTI	0
NEURONA	90
NEURONAS	0

MEROZOITE	634
((NG.IN. OR BIGBIE.IN. OR WHALEN.IN.) AND (SAROCOCYSTIS OR NEURONA OR MEROZOITE)).PGPB,USPT,USOC,EPAB,JPAB,DWPI,TDBD.	8

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20050260692. 13 Apr 05. 24 Nov 05. Positive selection in the immuno-dominant surface antigen 1 (SAG1) of sarcocystis neurona, agent of EPM: potential as a diagnostic reagent and as a component of a sub-unit vaccine. Mansfield, Linda S., et al. 435/7.22; G01N033/53 G01N033/569.

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☐ 2. 20030219381. 22 May 02. 27 Nov 03. Animal model for infection by an apicomplexan parasite. Ellison, Siobhan Patricia. 424/9.2; 435/258.1 A61K049/00 C12N001/10.

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☐ 3. 20020187517. 07 May 02. 12 Dec 02. Monoclonal antibodies to Sarcocystis neurona and uses therefor. Marsh, Antoinette. 435/7.22; 435/7.92 530/388.6 G01N033/53 G01N033/569 G01N033/537 G01N033/543 C07K016/20.

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☐ 4. 20020143018. 05 Mar 02. 03 Oct 02. Praziquantel compounds for treating diseases due to Sarcocystis, Neospora, Toxoplasma and Isospora. Kennedy, Thomas J.. 514/250; A61K031/4985.

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☐ 5. 20020041886. 23 Apr 01. 11 Apr 02. Equine protozoal myeloencephalitis vaccine. Bigbie, Rocky Barry, et al. 424/269.1; 435/258.1 514/44 A61K048/00 C12N001/10 A61K039/002.

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☐ 6. 6780415. 22 May 02; 24 Aug 04. Animal model for infection by an apicomplexan parasite. Ellison; Siobhan P.. 424/184.1; 424/265.1 424/269.1 424/272.1 424/273.1 435/440. A61K039/00 A61K039/002 A61K039/005 A61K039/015 C12N015/00 .

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☐ 7. 6489148. 12 May 00; 03 Dec 02. Immunoassay for equine protozoal myeloencephalitis in horses. Mansfield; Linda S., et al. 435/183; 435/34 435/7.1 435/7.2 435/7.22 436/518 530/388.6. C12N009/00 G01N033/55 G01N033/53 G01N033/567 C12Q001/04 .

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☐ 8. 6429211. 23 May 00; 06 Aug 02. Praziquantel compounds for treating diseases due to Sarcocystis Neospora Toxoplasma and Isospora. Kennedy; Thomas J.. 514/308; A61K031/47 .

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☐ 9. 6344337. 18 Feb 00; 05 Feb 02. Antigen test to detect equine protozoal myeloencephalitis in horse serum and cerebrospinal fluid. Mansfield; Linda S., et al. 435/7.2; 435/34 530/388.6. G01N033/53 .

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☐ 10. 6255308. 30 Apr 98; 03 Jul 01. Treatment of equine protozoal myeloencephalitis. Russell; Meri Charm, et al. 514/256; 514/275 514/601. A61K031/505 A61K031/18 .

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☐ 11. 6194408. 22 Dec 98; 27 Feb 01. Triazineone compounds for treating diseases due to Sarcocystis, Neospora and Toxoplasma. Kennedy; Thomas J.. 514/241; 514/242 514/243. A61K031/53 .

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☐ 12. 6153394. 18 Sep 98; 28 Nov 00. Immunoassay for equine protozoal myeloencephalitis in horses. Mansfield; Linda S., et al. 435/7.22; 435/7.1 436/518 436/523 436/528 436/543. G01N033/52 G01N033/569 G01N033/53 G01N033/543 G01N033/544 .

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☐ 13. 6150361. 22 Dec 98; 21 Nov 00. Triazineone compounds for treating diseases due to sarcosystis, neospora and toxoplasma. Kennedy; Thomas J.. 514/241; 514/242. A61K031/53 .

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☐ 14. 6110665. 14 Feb 95; 29 Aug 00. Sarcocystis neuronadiagnostic primer and its use in methods of equine protozoal myeloencephalitis diagnosis. Fenger; Clara K., et al. 435/6; 435/91.2 536/24.32 536/24.33. C12Q001/68 C12P019/34 C07H021/04 .

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☐ 15. 5883095. 07 Aug 97; 16 Mar 99. Formulations and methods to treat and prevent equine

protozoal myeloencephalitis. Granstrom; David, et al. 514/242; 514/241 514/275. A61K031/53 A61K031/505 .

☐ 16. 5830893. 23 Apr 97; 03 Nov 98. Treatment of equine protozoan myeloencephalitis using triazinediones. Russell; Meri Charmyne. 514/242; 514/557. A61K031/53 A61K031/495 A61K031/19 .

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Term	Documents
EQUINE	12682
EQUINES	1610
NEURONA	63
NEURONAS	0
((NEURONA.CLM.) AND (EQUINE.CLM.)).PGPB,USPT,USOC,EPAB,JPAB,DWPI,TDBD.	16
(EQUINE.CLM. AND NEURONA.CLM.)).PGPB,USPT,USOC,EPAB,JPAB,DWPI,TDBD.	16

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File: USPT

Feb 5, 2002

US-PAT-NO: 6344337

DOCUMENT-IDENTIFIER: US 6344337 B1

**\*\* See image for Certificate of Correction \*\***

TITLE: Antigen test to detect equine protozoal myeloencephalitis in horse serum and cerebrospinal fluid

DATE-ISSUED: February 5, 2002

## INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Mansfield; Linda S.	Bath	MI		
Rossano; Mary G.	Mason	MI		
Murphy; Alice J.	St. Johns	MI		
Vrable; Ruth A.	Williamston	MI		

US-CL-CURRENT: 435/7.2; 435/34, 530/388.6

## CLAIMS:

We claim:

1. In a method for detecting the presence of Sarcocystis neurona in an equine in an immunoassay, the improvement which comprises reacting a biological sample from the equine suspected of harboring the Sarcocystis neurona with at least one isolated antibody specific for a 16 (.-.4) kDa Sarcocystis neurona antigen and at least one isolated antibody specific for a 30 (.-.4) kDa Sarcocystis neurona antigen, wherein each antibody binds its respective antigen to form an antibody-antigen complex.

2. The method of claim 1, wherein the antibody-antigen complex is detected with a labeled antibody against the antigen or the antibody in the antibody-antigen complex.

3. The method of claim 2 wherein the label is selected from the group consisting of alkaline phosphatase, horseradish peroxidase, fluorescent compounds, luminescent compounds, colloidal gold, and magnetic particles.

4. The method of claim 2 wherein the label is biotin which is reacted with peroxidase conjugate and then detected by reaction with an appropriate color forming substrate.

5. The method of any one of claims 1, 2, 3, or 4 wherein the biological sample is selected from the group consisting of serum, cerebrospinal fluid, and cell culture fluid from equine dermal cells infected with Sarcocystis neurona from a biological sample.

6. The method of claim 1 wherein the antibody against the antigen is immobilized on a support selected from the group consisting of a membrane or a plate.

7. In a method for detecting the presence of *Sarcocystis neurona* in an equine in an immunoassay, the improvement which comprises reacting a biological sample from the equine suspected of harboring the *Sarcocystis neurona* with at least one monoclonal or isolated polyclonal antibody specific for a 16 (±.4) kDa *Sarcocystis neurona* antigen and at least one monoclonal or isolated polyclonal antibody specific for a 30 (±.4) kDa *Sarcocystis neurona* antigen, wherein each monoclonal antibody binds its respective antigen to form an antibody-antigen complex.

8. The method of claim 7 wherein the antibody-antigen complex is detected with a labeled antibody against the antigen or the antibody in the antibody-antigen complex.

9. The method of claim 8 wherein the label is selected from the group consisting of alkaline phosphatase, horseradish peroxidase, fluorescent compounds, luminescent compounds, colloidal gold, and magnetic particles.

10. The method of claim 8 wherein the label is biotin which is reacted with peroxidase conjugate and then detected by reaction with an appropriate color forming substrate.

11. The method of any one of claims 7, 8, 9, or 10 wherein the biological sample is selected from the group consisting of serum, cerebrospinal fluid, and cell culture fluid from equine dermal cells infected with a biological sample.

12. The method of claim 7 wherein the monoclonal antibody against the antigen is immobilized on a support selected from the group consisting of a membrane or a plate.

13. The method of claim 8 wherein the labeled antibody is a second monoclonal antibody.

14. A method for detecting *Sarcocystis neurona* in an immunoassay comprising:

(a) reacting a biological sample from an equine suspected of harboring the *Sarcocystis neurona* with at least one monoclonal antibody specific for a 16 (±.4) kDa *Sarcocystis neurona* antigen and at least one monoclonal antibody specific for a 30 (±.4) kDa *Sarcocystis neurona* antigen wherein each monoclonal antibody is immobilized on a support and wherein each monoclonal antibody binds its respective antigen to form a complex, and

(b) detecting the complex.

15. The method of claim 14 wherein the complex is detected by a labeled antibody against the antigen or monoclonal antibody in the complex.

16. The method of claim 15 wherein the labeled antibody is a second monoclonal antibody.

17. The method of claim 15 or 16 wherein the label is selected from the group consisting of alkaline phosphatase, horseradish peroxidase, fluorescent compounds, luminescent compounds, colloidal gold, and magnetic particles.

18. The method of claim 17 wherein the label is biotin which is reacted with



peroxidase conjugate and then detected by reaction with an appropriate color forming substrate.

19. The method of claim 14 wherein the biological sample is selected from the group consisting of serum, cerebrospinal fluid, and cell culture fluid from equine dermal cells infected with Sarcocystis neurona from a biological sample.

20. The method of claim 14 wherein the monoclonal antibody against the antigen is immobilized on the support selected from the group consisting of a membrane or a plate.

21. A kit for detecting Sarcocystis neurona in a biological sample from an equine comprising:

(a) at least one monoclonal or isolated polyclonal antibody against a 16 (±.4) kDa Sarcocystis neurona antigen and at least one monoclonal or isolated polyclonal antibody against a 30 (±.4) kDa Sarcocystis neurona antigen, wherein each antibody binds its respective antigen to form a complex;

(b) a positive control comprising the 16 (±.4) kDa Sarcocystis neurona antigen and a positive control comprising the 30 (±.4) kDa Sarcocystis neurona antigen; and

(c) a reagent for detecting the complex formed between the antibody and the Sarcocystis neurona antigen.

22. The kit of claim 21 wherein the reagent for detecting the complex consists of a labeled antibody against the antigen or antibody in the complex.

23. The kit of claim 22 wherein the label is selected from the group consisting of alkaline phosphatase, horseradish peroxidase, fluorescent compounds, luminescent compounds, colloidal gold, and magnetic particles.

24. The kit of claim 23 wherein the label is biotin which is reacted with peroxidase conjugate and then detected by reaction with an appropriate color forming substrate.

25. The kit of claim 24 wherein the biological sample is from the group consisting of serum, cerebrospinal fluid, and cell culture fluid from equine dermal cells infected with Sarcocystis neurona from a biological sample.

26. The kit of claim 21 wherein the monoclonal antibody against the antigen is immobilized on a support selected from the group consisting of a membrane or a plate.

27. The kit of claim 22 wherein the labeled antibody is a second monoclonal antibody.

28. A kit for the detection of disease caused by Sarcocystis neurona in an equine which comprises:

(a) a support with a monoclonal antibody against a first epitope of a 16 (±.4) kDa Sarcocystis neurona antigen and a monoclonal antibody against a first epitope of a 30 (±.4) kDa antigen immobilized on a surface of the support to bind the antigen in a biological sample from the equine;

(b) a first labeled monoclonal antibody against a second epitope of the 16 (.-.4) kDa antigen and a second labeled monoclonal antibody against a second epitope of the 30 (.-.4) kDa antigen to bind the antigen bound by the monoclonal antibody immobilized on the membrane; and

(c) a reagent for detection of the first labeled monoclonal antibody bound to the 16 (.-.4) kDa antigen and a reagent for detection of the second labeled monoclonal antibody bound to the 30 (.-.4) kDa antigen.

29. The kit of claim 28 wherein the label is selected from the group consisting of alkaline phosphatase, horseradish peroxidase, fluorescent compounds, luminescent compounds, colloidal gold, and magnetic particles.

30. The kit of claim 29 wherein the label is biotin which is reacted with peroxidase conjugate and then detected by reaction with an appropriate color forming substrate.

31. The kit of claim 28 wherein the biological sample is selected from the group consisting of serum, cerebrospinal fluid, and cell culture fluid from equine dermal cells infected with Sarcocystis neurona from a biological sample.

32. The method of claim 28 wherein the monoclonal antibody against the antigen is immobilized on the support selected from the group consisting of a membrane or a plate.

33. A monoclonal antibody against a 16 (.-.4) kDa antigen of Sarcocystis neurona.

34. A monoclonal antibody against a 30 (.-.4) kDa antigen of Sarcocystis neurona.

35. An isolated DNA encoding a 16 (.-.4) kDa antigen of Sarcocystis neurona.

36. An isolated DNA encoding a 30 (.-.4) kDa antigen of Sarcocystisneurona.